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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
10/065,610	11/04/2002	Chi-Ming Chen	9668-US-PA	2460

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JIANQ CHYUN INTELLECTUAL PROPERTY OFFICE
7 FLOOR-1, NO. 100
ROOSEVELT ROAD, SECTION 2
TAIPEI, 100
TAIWAN

EXAMINER	
WOOLWINE, SAMUEL C	
ART UNIT	**PAPER NUMBER**
1637	

DATE MAILED: 02/02/2006

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/065,610	CHEN, CHI-MING	
	Examiner	Art Unit	
	Samuel Woolwine	1637	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *12 December 2005*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-6, 8 and 16* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-6,8 and 16* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date 12/13/05
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: _____.

DETAILED ACTION

The amendments to claim 1, addition of claim 16, and the cancellation of claims 7 and 9-15 in the response filed on 12/12/2005 is acknowledged. Claims 1-6, 8 and 16 are pending in the instant application.

Response to arguments-Claim objections

As a result of the cancellation of claim 15 in Applicant's response filed on 12/12/2005, the objection to claim 15 stated in the previous Office Action entered 9/13/2005 is withdrawn.

Response to arguments-Drawings

As a result of substitute drawings submitted with Applicant's response filed on 12/12/2005, the objection to the drawings stated in the previous Office Action entered 9/13/2005 is withdrawn.

Response to arguments: Claim Rejections - 35 USC § 112 Written Description

As a result of the claim amendments entered in Applicant's response filed on 12/12/2005, the rejection of claims 1-8 stated in paragraph 8, page 3 of the previous Office Action entered 9/13/2005 is withdrawn as no longer applicable.

Claim Rejections - 35 USC § 112 Enablement

Claims 1-6, 8 and 16 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enablement requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention.

Factors to be considered in determining whether a disclosure meets the enablement requirement of 35 USC 112, first paragraph, have been described by the court in *In re Wands*, 8 USPQ2d 1400 (CAFC 1988). *Wands* states at page 1404,

> "Factors to be considered in determining whether a disclosure would require undue experimentation have been summarized by the board in Ex parte Forman. They include (1) the quantity of experimentation necessary, (2) the amount of direction or guidance presented, (3) the presence or absence of working examples, (4) the nature of the invention, (5) the state of the prior art, (6) the relative skill of those in the art, (7) the predictability or unpredictability of the art, and (8) the breadth of the claims."

The nature of the invention

The claims are drawn to methods to determine the sequence of a nucleic acid by controlling its rate of translocation through a synthetic thin film nanopore, and measuring the fluctuations in the electrical current thus produced. The invention is in a class of invention which the CAFC has characterized as "the unpredictable arts such as chemistry and biology." Mycogen Plant Sci., Inc. v. Monsanto Co., 243 F.3d 1316, 1330 (Fed. Cir. 2001).

The breadth of the claims

The claims are broadly drawn to methods for determining the sequence of nucleic acids using a thin film nanopore. These claims would therefore encompass DNA and RNA, single- or double-stranded, including any modifications such as 7-methylguanosine-capped mRNA, methylated DNA, or the various modified residues

found in tRNA. The specification does not describe how the claimed methods would allow one to determine the identities of such modifications. With particular regard to double-stranded nucleic acids, the basis described in the specification for discriminating among the 4 bases normally found in nucleic acids (i.e. the unique blockage current signature of each base) would only allow one to determine the sequence of single-stranded nucleic acids. This is because, while the blockage current signature for a GC base pair may be distinguished from that of an AT base pair, one would not be able to deduce which strand of a double-stranded nucleic acid molecule contained G and which contained C, for example.

Quantity of experimentation

The quantity of experimentation required by one of ordinary skill in the art to practice the methods claimed by Applicant is large. This is mainly due to the fact that the methods have not been reduced to practice and are based solely on predictions involving computer simulations. Applicant has given no indication of actually fabricating the device required to practice the method. A publication by Applicant subsequent to the filing of the instant application also provides no indication of a working device for practicing the methods claimed (Chen & Peng, 2003). The Court in In re Ghiron, 442 F.2d 985, 991, 169 USPQ 723, 727 (CCPA 1971), made clear that if the practice of a method requires a particular apparatus, the application must provide a sufficient disclosure of the apparatus if the apparatus is not readily available. While Applicant describes the essential features of such an apparatus in the specification, the fabrication of such a device is not described in the specification in such detail as to

obviate undue experimentation by one of ordinary skill in the art. The following paragraph discusses some features of the apparatus required to practice the claimed methods that are unpredictable and would therefore require undue experimentation for reduction to practice.

The unpredictability of the art and the state of the prior art

The current state of the art indicates that a great deal of further experimentation and inventiveness would be required to implement the methods claimed by Applicant. For instance, Aksimentiev et al. (2004) states:

> "For synthetic nanopores observed blockades presently show irregular shapes (see Fig. 1) due to fluctuations of the reduced current within one blockade event; some blockades have a positive spike of the ionic current above the open pore level at the end of the blockade" (p. 2087, column 1, lines 7-11).

Aksimentiev et al. (2004) cites Applicant's 2003 publication describing the methods of the instant application, and was therefore aware of Applicant's invention, and yet states:

> "The manufacturing of actual nanopores and electrical recordings are still at an early stage that needs guidance toward optimal strategies toward the ultimate goal of sequencing DNA" (p. 2095, column 2, lines 29-32).

Applicant refers to Li et al. (2001) for a method of forming the nanopore to be used in the method claimed in the instant application. However, the nanopores fabricated by Li et al. (2001) are approximately 10nm in length, a distance which can accommodate 10-15 nucleotides at a time. Therefore, the blockage current measured would be the result of the presence of multiple nucleotides in the pore simultaneously and could not be used to identify the sequence of the nucleic acid molecule. This technical challenge (i.e. the length of the pore channel) has still not been completely met, as stated by Heng et al. (2004):

> "...the scaling of MOS [metal oxide semiconductor] field effect transistor (MOSFET) designs to nanometer-scale gate length (Sorsch et al., 1998) have produced high-integrity, ultrathin films as thin as 0.7 nm (which is comparable to the spacing between basepairs in DNA) that are suitable for membranes. By combining these two elements, we have already produced ~1-nm diameter pores that are smaller in diameter than DNA in solution, **through membranes ~5-50-nm thick**" (p. 2905, column 2, lines 22-29, emphasis added).

Therefore, even as late as 2004, the technical hurdle of producing a nanopore in a film thin enough to enable single nucleotide resolution based on blockage current has still not been overcome.

An additional issue of unpredictability that the specification does not address is the potential for interaction of the nucleic acid with the material of the nanopore. Aksimentiev et al. (2004) states:

> "A strong hydrophobic interaction of the DNA bases with the surface of the pore (Fig. 7) favors an unzipped conformation of a double-stranded DNA inside the pore (Figs. 6 and 9). A significant reduction of the ionic current can be observed even when DNA is not transiting the pore (Fig. 8), such that only part of the ionic current blockade measured experimentally reflects actual DNA translocation events" (p. 2095, column 2, lines 19-26).

The issue of nucleic acid interacting with the pore is also raised by Heng et al. (2004):

> "With or without deconvolution, it is evident from the variety of transients observed that the level of the blocking current changes during the time interval of the transient, which we interpret as the molecule interacting with the pore. Molecular dynamics simulations described below and in (Aksimentiev et al., 2004), indicate that the level of the blocking current is correlated to the velocity of the molecule in the pore and with the bulk electrolyte flow accompanying the translocation event. Consequently, we expect that interactions between the molecule, the pore, and the membrane give rise to a nonuniform molecular velocity and bulk electrolyte flow, which are manifested as variations in the blocking current during a single translocation. Moreover, the simulations show that when DNA exits the pore, ions accumulating near the mouth are also released resulting in the positive current spike similar to that observed experimentally on the rising edge of the transient in Fig. 3" (p. 2908, column 1, line 23 through column 2, line 7).

Yet another unpredictable feature of the method claimed by Applicant is the effect of the rotating electric field on the perpendicular electric field required both for the translocation of the nucleic acid through the nanopore as well as the electronic signal allowing the identification of each nucleotide passing through the nanopore. At best, the

rotating electric field would have no effect on the perpendicular electric field or the electric current passing through the nanopore. At worst, the perpendicular electric field would experience such interference from the rotating electric field that the nucleic acid might not translocate at all, or the electric current passing through the nanopore would be so noisy as to render identification of the translocating nucleic acid sequence impossible. Without actual reduction to practice, it is not possible to predict with reasonable certainty whether the claimed methods would work.

Working examples

As Applicant's invention has not been reduced to practice, there are no working examples provided in the specification.

Guidance in the specification

The specification provides guidance as to the essential features of the device required for practicing the method, but provides little guidance as to the actual fabrication of such a device. The specification does offer general guidance for optimizing parameters such as the applied voltage, the frequency of the rotating electric field, the use of nucleic acid "tags" to attach to the ends of the nucleic acid molecule to be sequenced, and the modification of certain bases to create more distinction in the blockage current signal.

Level of skill in the art

The level of skill in the art is deemed to be high.

Conclusion

In the instant application, a method for determining the sequence of nucleic acids is proposed in which one electric field is applied perpendicular to a thin film containing a single synthetic thin film nanopore in order to drive a nucleic acid molecule through the nanopore, while a second rotating electric field controls the rate of translocation of the nucleic acid through the nanopore, thus allowing the change in the electric current through the nanopore to be used as a signature for the identification of each nucleotide as it passes through. While the methods claimed by Applicant might allow for the controlled rate of translocation of the nucleic acid molecule through the nanopore, the likelihood that one of skill in the art could readily practice the methods claimed by Applicant is remote given:

I. the method has not been reduced to practice

II. the device required for practice of the methods has not been fabricated

III. the methods are based on computer simulations that may not adequately reflect all of the unpredictable realities of the system under investigation (i.e. the movements of polynucleotides in solution, the interaction of polynucleotides with the materials of the nanopore, the effects of the rotating electric field on the electronic signals of the nanopore)

IV. the limitations in the current state of the art (chiefly, the lack of a nanopore-containing film thin enough to achieve single nucleotide resolution)

It would therefore require a person of ordinary skill in the art undue experimentation to practice the methods claimed in the instant application. Therefore, rejection of claims 1-15 under 35 U.S.C. 112, first paragraph is proper.

Response to arguments: Claim Rejections - 35 USC § 112 Enablement

Applicant's arguments regarding the enablement rejection have been carefully considered but are not found persuasive.

With regard to the amendments to claim 1, Applicant argues that "amended claim 1 is directed to the method for controlling a nucleic acid sequence, instead of a nucleic acid sequencing method" (page 9 of arguments). However, amended claim 1 contains the limitation "suitable for determining a sequence of the nucleic acid sequence". Therefore, the claims still suffer from a lack of enablement for the same reasons stated in the enablement rejection above.

With regard to *DeGeorge v. Bernier*, the issue in question of before the Court was whether the DeGeorge disclosure was enabling despite the contention by Bernier that "the control circuits for the connections and coils in the TCCPI circuit include contacts operated by coils in the word processor and the control circuits include connections with signals from the word processor that are not identified". The Court held DeGeorge's disclosure to be enabled, in part as a result of expert testimony "that 'any logic designer of a normal ability should be able to implement functions given this much description [in the per se '670 disclosure] about them.'"

The question raised by the Examiner concerning the instant application is not whether the two electric fields of the method are adequately described, but whether the electric field required for electrophoretic movement through the nanopore would interfere with the rotating electric field required for electrorotation, and vice versa. This question was raised by the Examiner because the invention of the instant application has not yet been reduced to practice, and a further search by the Examiner for examples of these two types of electric field in simultaneous operation did not resolve the question. Chapter 2145 of the MPEP states: "The arguments of counsel cannot take the place of evidence in the record. *In re Schulze*, 346 F.2d 600, 602, 145 USPQ 716, 718 (CCPA 1965)". An affidavit from a qualified expert, for example, an electrical engineer, or some other evidentiary material, such as data generated by a working device, would be more persuasive in settling this particular issue of unpredictability.

Regarding the issues of "quantity of experimentation" on page 10 of Applicant's arguments filed on 12/12/2005, the amended claims still contain the limitation "suitable for determining a sequence of the nucleic acid sequence". The issue of undue experimentation is not directed to the process of electrorotation *per se*, for which a great deal of literature may be found in the prior art, but for the integration of electrorotation and electrophoretic movement of nucleic acid through a nanopore as well as for the fabrication of said nanopore thin film, which must be thin enough to allow for single nucleotide resolution in order to be "suitable for determining a sequence of the nucleic acid sequence".

Regarding the "working examples" referred to on pages 10 and 11 of Applicant's arguments, it is noted that these are not true working examples of the invention, but computer simulations *predicting* what would occur *if* the invention were actually built and used.

Regarding the acceptability of non-working embodiments alluded to in *Decca, Ltd. v. United States*, Applicant's point is well-taken. However, a disclosure must be enabling for at least *one* working embodiment, and the disclosure of the instant application is not so enabling. A significant defect of the disclosure of the instant application is the lack of direction for the fabrication of a nanopore film thin enough to accomplish the purpose of the claimed invention, which must be "suitable for determining a sequence" and allow the measuring of "a blockage current of each of the nucleotides" (claim 1 of the instant application as currently amended). This deficiency in the state of the art at the time the application was filed, and indeed in the current state of the art, was a major point raised in the enablement rejection and was not addressed at all in Applicant's response.

The rejection of claims 1-6, 8 and 16 under 35 U.S.C. 112, 1st paragraph is deemed proper. Accordingly this rejection is made FINAL.

Conclusion

No claims are allowed. This action is made FINAL.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Samuel C. Woolwine whose telephone number is (571) 272-1144. The examiner can normally be reached on 8:30-5:00.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on (571) 272-0782. The fax phone number for the organization where this application or proceeding is assigned is 703-872-9306. Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

JEFFREY FREDMAN
PRIMARY EXAMINER
1/3/06

scw